AD_____

Award Number: DAMD17-99-1-9062

TITLE: DNA Damage, Fruits and Vegetables and Breast Cancer Prevention

PRINCIPAL INVESTIGATOR: Henry Thompson, Ph.D.

CONTRACTING ORGANIZATION: AMC Cancer Research Center
Denver, Colorado 80214

REPORT DATE: August 2000

TYPE OF REPORT: Annual

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

DISTRIBUTION STATEMENT: Approved for Public Release;
Distribution Unlimited

The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army position, policy or decision unless so designated by other documentation.

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 074-0188

1. AGENCY USE ONLY (Leave blank)	2. REPORT DATE	3. REPORT TYPE AND DATES COVERED
	August 2000	Annual (1 Aug 99 – 31 Jul 00)

4. TITLE AND SUBTITLE
DNA Damage, Fruits and Vegetables and Breast Cancer Prevention

5. FUNDING NUMBERS
DAMD17-99-1-9062

6. AUTHOR(S)
Henry Thompson, Ph.D.

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)
AMC Cancer Research Center
Denver, Colorado 80214

E-MAIL:
thompsonh@amc.org

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)
U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

10. SPONSORING / MONITORING AGENCY REPORT NUMBER

11. SUPPLEMENTARY NOTES

12a. DISTRIBUTION / AVAILABILITY STATEMENT
Approved for public release; distribution unlimited

12b. DISTRIBUTION CODE

13. ABSTRACT ***(Maximum 200 Words)***
The purpose of this project is to evaluate the effect(s) of increasing fruit and vegetable intake on oxidative DNA damage and lipid peroxidation in a population of women at elevated risk for breast cancer. The rationale that underlies the work proposed is based on evidence that the occurrence of DNA mutations are essential steps in carcinogenesis and that these mutagenic events can result from oxidative stress, even in the absence of exogenous carcinogens. The effects of consuming a recipe-defined diet designed to provide three (control) or ten (intervention) servings of fruits and vegetables per day for a period of 8 weeks on measures of oxidative damage to DNA and lipids is being determined. During this reporting period, we have expanded the initial two-week dietary intervention so that subjects can follow the dietary prescription for 8 weeks. Based on study results, key variables that required modification were related to convenience of meal preparation and flexibility in the selection of alternative foods from within the same botanical family. Barriers research identified alienation from family members due to eating a different diet, limitations in eating meals out of home, and lack of favorite food items as significant obstacles to maintaining dietary compliance.

14. SUBJECT TERMS	15. NUMBER OF PAGES
Breast Cancer, Oxidative Damage, Vegetablles, Fruit	53
	16. PRICE CODE

17. SECURITY CLASSIFICATION OF REPORT	18. SECURITY CLASSIFICATION OF THIS PAGE	19. SECURITY CLASSIFICATION OF ABSTRACT	20. LIMITATION OF ABSTRACT
Unclassified	Unclassified	Unclassified	Unlimited

NSN 7540-01-280-5500

Standard Form 298 (Rev. 2-89)
Prescribed by ANSI Std. Z39-18
298-102

Table of Contents

Introduction

The objective of this research project is to determine the effect(s) of increasing fruit and vegetable intake on oxidative DNA base damage and lipid peroxidation in a population of women at elevated risk for breast cancer. The rationale that underlies the work proposed is based on evidence that the occurrence of DNA mutations are essential steps in carcinogenesis and that these mutagenic events can result from oxidative stress, even in the absence of exogenous carcinogens. The effects of consuming a recipe-defined diet designed to provide three (control) or ten (intervention) servings of fruits and vegetables per day for a period of 8 weeks on measures of oxidative damage to DNA and lipids is being determined. Urine and blood components are being assessed for oxidative endpoints and plasma is being evaluated for biochemical markers of edible plant consumption.

Body

1. Approved Statement of Work

During the negotiations involved in the award of this grant, reductions in the scope of the originally proposed work plan were implemented based on the review group's critique of the application. Accompanying reductions in the budget were also made. The final approved Statement of Work is as follows:

To test whether an increase in consumption of fruits and vegetables will decrease indicators of oxidative cellular damage in women at high risk for breast cancer occurrence or reoccurrence.

a. Initiate recruitment 2 months prior to initiation of a study group into the investigation (2 months).
b. Conduct intervention (two-week run in and 8 week intervention) in a total of 2 study groups (50 subjects, 25/group) (2.5 months).
c. Perform laboratory analyses (6 months for a study group of 50 subjects).
d. Evaluate results (2 month per study group).
e. Repeat steps a-d an additional three times (Years 1-3). We anticipate that recruitment will be completed during year three, and that laboratory and statistical analyses will continue throughout the project.
f. Summarize results and write reports and manuscripts (Years 1-3).

2. Project Initiation

As noted in the original application, this project was based on pilot work in which we studied the effects of a two-week recipe defined diet on oxidative markers. Upon commencement of work on this project, a multi-pronged plan of attack was implemented. Its elements included: 1) modification of the recipe-defined menus for use in an 8-week intervention study; 2) development and testing of intervention materials; and 3) further evaluation of the candidate oxidative markers. Progress on each area is reported in subsequent sections of this report.

3. Modification of the Recipe Defined Menus

a. Focus group analysis of two-week dietary intervention program

Subjects that had participated in previous two-week dietary intervention studies were invited to attend focus group meetings to elicit their comments and suggestions on the menus, recipes, and other aspects of the dietary intervention in which they had participated. Open-ended questions were used to promote discussion of the aspects of the two-week dietary intervention that needed to be changed if subjects were to follow the intervention for a period of 8 weeks. The following barriers were identified: 1) the amount of time required for "in home" meal preparation including the weighing of all food items, i.e. lack of convenience, 2) difficulty of eating meals out of home, 3) inability to follow the diet while traveling and during normal business activities,

e.g. business lunches and dinners, 4) exclusion of "favorite foods", 5) meal repetition, 6) limitations on the consumption of alcoholic beverages, 7) prohibition of nutritional supplement use.

b. Modification of the dietary intervention program

Based on the results of the focus groups, the project staff defined four elements of the intervention program for discussion and analysis. They were: convenience, flexibility, choice, limitations.

Convenience

The primary barrier identified in the focus groups was lack of convenience. To address this issue several strategies were evaluated. They included: 1) the use of convenience foods that could be purchased from local grocery stores, 2) the use of a feeding study approach in which the majority of the meals would be provided to subjects, 3) the identification of the amounts and types of vegetables and fruit to be consumed each day without further specification of foods to be eaten, 4) the development of convenience foods for use in the study. Given the significance of this barrier, considerable time and effort was committed to evaluating these alternatives. The results of those deliberations can be summarized as follows.

The use of convenience foods that could be purchased from local grocery stores This approach is deemed to have considerable merit relative to the ultimate translatability of the intervention strategy to large segments of the population. However, the majority of the convenience foods provided as meals contain limited amounts of vegetables and almost no fruit, the variety of vegetables and fruit are limited, and such products are expensive, can be high in calories, and their quality control is unknown. For these reasons, this approach was not further pursued.

The use of a feeding study approach in which the majority of the meals would be provided to subjects This approach was deemed desirable because of the control that it would provide of the foods consumed. However, this approach has shortcomings for this study population. The population has a mean age of 48,and the majority of women are employed full time. This approach would be likely to exclude the possibility of many women to participate, and they would not learn the skills necessary to translate the principles of the intervention into their daily lives. This intervention would also be expensive to implement. For these reasons, this approach was not further pursued.

T*he identification of the amounts and types of vegetables and fruit to be consumed each day without further specification of foods to be eaten.* Ultimately, this approach may offer the greatest opportunity for translation; however, at this stage of hypothesis testing, the loss of control over other aspects of the diet that is inherent in this approach was considered unacceptable. For this reason, this approach was not further pursued.

The development of convenience foods for use in the study. This approach was considered to offer the greatest opportunity for successful implementation because it would allow the use of many of the recipes developed for the original diet while keeping most elements of the menu plan intact. Moreover, this approach also addressed the concerns identified when the use of convenience foods was considered (see above). To pursue this approach, local grocery chains that prepare entrees on site were identified and discussions of interest and feasibility were initiated. The Wild Oats foodchain expressed interest and had staff that were headed by a certified chef with considerable experience in the conversion of recipes for individuals for quantity production. For reasons of feasibility, the ability to prepare all recipes and provide them in a "frozen format" was deemed essential. Further aspects of this activity are presented under "Intervention material development and testing".

The other major change made was to switch from the use of scales to weigh all foods to the use of standard household measures, i.e. cups.

Flexibility

In our analysis of the focus group data, a number of barriers identified were related to the lack of flexibility in the ability to accommodate business activities, trips, and other personal activities, all of which necessitate the

eating of meals out of the home environment. Further discussions with former study participants indicated that one solution that would address this situation would be to allow two meals per week to be "free meals", i.e. not prescribed by the menu plan. It was decided that this approach should be incorporated into the dietary intervention plan. However, we decided that at this stage of hypothesis testing, that the need for out-of-town travel for more than two days during the intervention would serve as a basis for non inclusion in the study. However, it was also agreed that this decision be regularly scrutinized.

Choice

Suggestions for the incorporation of new food items into the diet were considered in conjunction with the repetitive use of certain food items. Menus were modified accordingly. For example, there was considerable interest in the inclusion of chocolate which was added in equal amounts to both the low and high vegetable and fruit diets. However, the decision to use a two week menu plan which repeated 4 times was considered important to hypothesis testing and this element of experimental design was retained.

Limitations

In concert with the focus group results, we decided to permit the consumption of red wine (or grape juice) on a daily basis. However, given the fact that the menu plan met or exceeded the RDA's for all nutrients, we affirmed the decision to require that participants refrain for taking vitamin and mineral supplements.

4. Development and evaluation of modified dietary intervention

a. Development of intervention materials

In order to incorporate the use of our previously developed recipes for provision in frozen form by a local grocery chain, the following process was used. The menu plan was reviewed for recipes that would be satisfactory when frozen. Our goal was to identify about one-half the food selections that could be provided to study participants in a convenience format. Once this task was successfully completed, the following approach was used for quantity food preparation:

1. Convert recipe from gram weights to cup measures
2. Resize recipes (now in cup measures) from 4 servings to 50 servings
3. Determine serving sizes for 4 calorie levels for each recipe
4. Wild Oats tests recipes at food service quantity portions (50 servings)
5. "Focus group" taste tests
6. Recipe revision, as needed (needed to adjust spices alot of times)
7. Wild Oats retesting of recipes, as needed

The cook book which is the primary intervention tool and that is specifically tailored to each individual's caloric requirements was then completely revised to reflect these changes. An example of one day's menu is Appendix A.

b. Evaluation of the modified intervention

In order to evaluate the suitability and feasibility of the modified menu plan, 40 subjects were recruited and randomly assigned to either the low or high vegetable and fruit intervention groups. The primary goal was for study participants to identify problems in the menus and intervention approach that would inhibit their ability to follow the menu plan for a total of eight weeks. The questionnaire that is Appendix B was developed and used as a component of the analysis plan.

Thirty-five subjects completed the study. The evaluation of study results is broken into the four areas that we outlined in our effort to overcome initially identified barriers.

Convenience

The use of the local grocery store to provide frozen entrée's was deemed highly desirable by study participants. Subjects were able to successfully obtain all entrees on a weekly basis without significant problems. No confusion of foods between the two study groups occurred. Moreover, the grocery store personnel found that

the system worked well, did not disrupt their normal activities, and the management and staff were excited to participate in the project.

Flexibility Subjects indicated that the ability to have two "free meals" a week provided adequate flexibility such that longer term adherence to the menu plan would be possible.

Choice
Relative to the acceptability of food items and the variety of foods in the menus, significant and extensive feedback was obtained. The net effect of the feedback was that the cookbook still required substantial modifications. As discussed below, these modifications were implemented and evaluated in a subsequent focus group.

Limitations
The ability to consume a glass of wine or grape juice was considered acceptable by the majority of study participants. It was determined that extensive discussion of the nutritional adequacy of diets addressed the majority of concerns about the use of supplements was satisfied. However, it was also determined that because of the health opinions of some individuals, that prohibition of supplement use is a key issue to discuss during subject recruitment.

c. Further modification of the intervention cookbook

Key issues that were identified were the need for modification of the recipes for the "convenience entrees", and the need for greater variation of and flexibility in the selection of foods to avoid dropout during an eight week intervention. Based on the suggestions obtained, recipes were modified and retested, and a new approach to increase flexibility was developed. In order to increase food choices, we used the principle that has been an underlying guide to vegetable and fruit selection for the menu plan, i.e. that food items from the same botanical family generally have more similar chemical composition than those in different botanical families. An exchange list for the "non grocery store supplied convenience items". For vegetable or fruit selections, subjects will given choices, but the choices are limited to items from the same botanical family. While we do lose some control over dietary composition, the evaluation of this approach was given enthusiastic support during focus group analysis. An example of this modified approach is provided as Appendix C. Other materials developed for the project are provided in Appendix D.

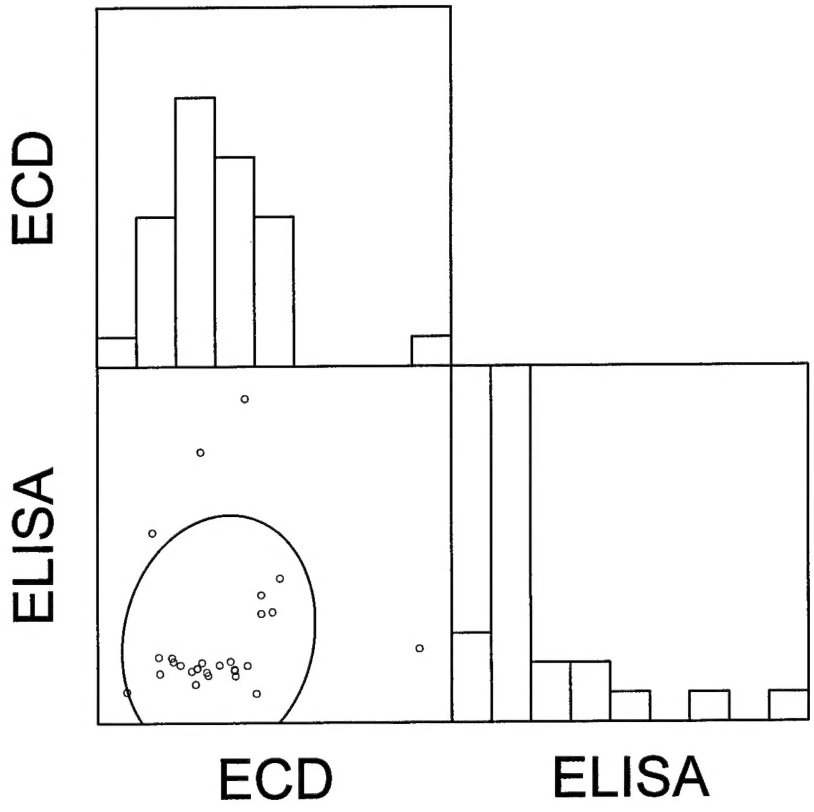
5. **Subject recruitment** With the completion of the final modifications of the cookbook, we began recruiting for study. During the months of July and August, 2000, sixty subjects have been recruited. The actual intervention will be initiated during September, 2000.

6. Investigation of Oxidative Markers
During the development of the dietary intervention materials, work has been ongoing relative to the markers of oxidative damage that will be assessed during the intervention study. Our progress is summarized in the following paragraphs.

a. **Assessment of urinary 8-OHdG** As noted in the grant application, we had some concern about the specificity of the ELISA assay for measuring urinary 8-OHdG. Therefore, we decided to compare results obtained using the ELISA method with results obtained by dual column high pressure liquid chromatography with electrochemical detection (ECD). We selected 28 samples for these analyses that were available in large amounts. The results are summarized in the following table.

	ECD	ELISA
N of cases	28	28
Minimum	1.664	1.195
Maximum	7.352	180.585
Mean	3.449	35.447
Standard Dev	1.078	42.676
C.V.	0.313	1.204

These data confirm our preliminary concern. Values determined by ECD are 10-fold lower and significantly less variable than those obtained by ELISA analysis. Moreover, as shown in the following graph, the correlation between these data was low, r^2=0.11, p=0.5 and not statistically significant. Also note the non gaussian distribution of the ELISA data. For these reasons, urinary 8-OHdG will be evaluated by dual column chromatograph with ECD.



The following paragraphs summarize our current thinking about the assessment of urinary 8-OHdG. We are considering publication of this information.

The limitations of urinary 8-OHdG as a marker for oxidative DNA damage, particularly by use of the Genox ELISA technology, are becoming obvious as research continues. Foremost among concerns about this marker is its uncertain origin. The appeal of urinary 8-OHdG stems from evidence that it occurs in urine after excision from nuclear DNA by repair processes [1], but some portion of excreted 8-OHdG may arise from mitochondrial DNA, cell turnover, or hydrolysis of oxidized deoxyguanosine triphosphate [2-4]. The suggestion that DNA repair contributes significantly to urinary 8-OHdG abundance is questionable, given what is known about DNA repair. It is generally accepted that non-bulky lesions such as 8-OHdG are removed from DNA by base excision repair (BER), as opposed to nucleotide excision repair (NER) [5;6]. The free oxidized base 8-hydroxyguanine (8-OHGua), not 8-OHdG, is the repair product one would expect from the glycosylase activity that is characteristic of BER. Although evidence for human endonuclease activity specific for 8-hydoxyguanine and without associated glycosylase activity has been reported [7], and such activity may be expected to generate 8-OHdG as a product, no evidence for this novel human endonuclease has been demonstrated *in vivo*. 8-OHdG would also be an expected product of nucleotide excision repair, and it has been proposed that NER can remove oxidized guanine from DNA under some conditions [8;9]. However, recent evidence indicates that BER accounts for the vast majority of excision from DNA of oxidized guanine bases, both transcription coupled and in non-transcribed DNA, in mammalian cells [9-11]. This does not necessarily mean that urinary 8-OHdG is without merit as a marker for oxidative damage, but that it is likely not a significant DNA repair product.

Urinary 8-OHdG that originates from mitochondrial DNA, cell turnover, or hydrolysis of oxidized deoxyguanosine triphosphate may still provide useful information on oxidative stress with mutagenic potential[1]. In addition to these concerns, the credibility of urinary 8-OHdG results obtained by use of the Genox assay has been called into question. The assay has been criticized for a lack of specificity and high variability, and we have generated data that is consistent with these shortcomings[12;13]. Analysis of urinary 8-OHGua, the logical choice as a urinary oxidative DNA damage marker given what is known about repair, is complicated by the substantial contribution from dietary sources to urinary 8-OHGua.

Despite the aforementioned limitations, both 8-OHdG and 8-OHG may be informative urinary oxidative indices. 8-OHGua abundance may be a useful marker for oxidative DNA damage if the dietary contribution is minimized and controlled for, and 8-OHdG, while not a DNA repair product per se, may still reflect oxidative stress. We are therefore investigating the use of multicolumn HPLC with electrochemical array detection for dependable analysis of both analytes [14].

b. Assessment of DNA oxidation by the "comet assay"

While there is a strong rationale for the assessment of oxidative DNA damage in lymphocytes via measurement of 8-OHdG, as detailed in the grant application, we recognize that one limitation of this approach is that 8-OHdG represents only one of many DNA oxidation products. Thus, it would be of value to the project to have additional assays of oxidative DNA damage that would complement the analysis of lymphocyte 8-OHdG. For this reason we have evaluated the relationship between lymphocyte 8-OHdG determined electrochemically and the assessment of DNA damage by the comet assay in the absence and presence of two enzymes (endonuclease-III and FPG). This application of the comet assay permits the estimation of DNA single strand breaks, and pyrimidine and purine oxidation. The following data summarize our findings.

Pearson correlation matrix

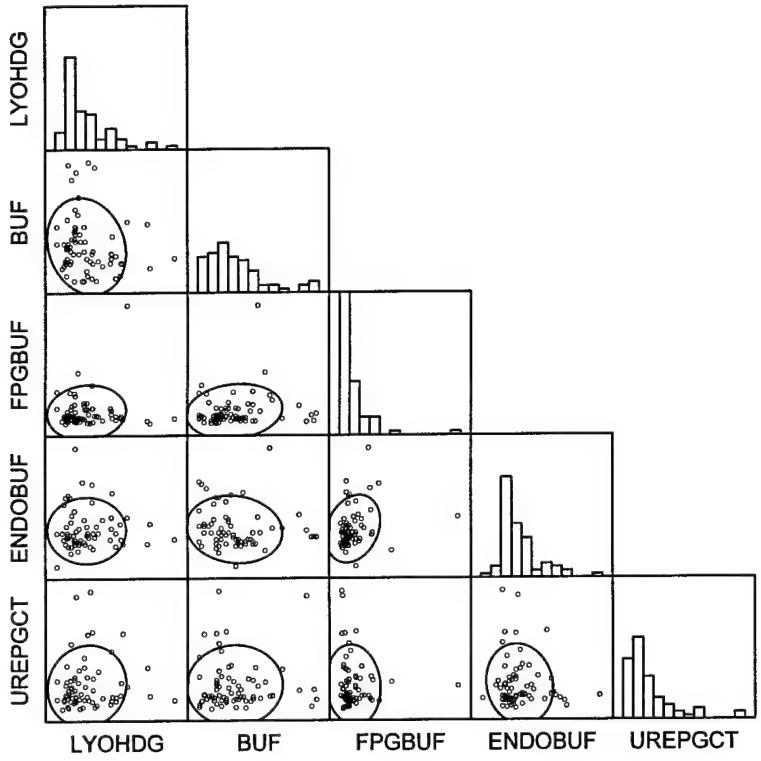
	LYOHDG	BUF	FPGBUF	ENDOBUF	UREPGCT
LYOHDG	1.000				
BUF	-0.163	1.000			
FPGBUF	0.071	0.123	1.000		
ENDOBUF	-0.005	-0.107	0.234	1.000	
UREPGCT	0.064	0.016	-0.081	-0.097	1.000

N for each assay=70.
Bartlett Chi-square statistic: 9.552 df=10 Prob= 0.481

Matrix of Bonferroni Probabilities

	LYOHDG	BUF	FPGBUF	ENDOBUF	UREPGCT
LYOHDG	0.000				
BUF	1.000	0.000			
FPGBUF	1.000	1.000	0.000		
ENDOBUF	1.000	1.000	0.511	0.000	
UREPGCT	1.000	1.000	1.000	1.000	0.000

Abbreviations: Lymphocyte 8-OHdG, LYOHDG; alkaline labile single strand breaks, BUF; oxidized purines, FPGBUF; oxidized pyrimidines, ENDOBUF; urinary 8-isoprostane F2-alpha, UREPGCT.



No statistically significant relationship was observed among these oxidative indices. Given that there is an extensive literature that documents the validity of each of these markers, as an index of damage to DNA or lipid, we conclude that each index is contributing information that is unrelated to the information provided by the other markers. Thus, we anticipate that the assessment of all markers is likely to give us the most complete picture of the changes in oxidative status induced by the dietary intervention. To our knowledge, a similar comparison of multiple markers has not been reported in the literature. Given the large N that these data represent, we are considering the preparation of these data for publication.

c. Preliminary analysis of the assessment of protein oxidation

During this reporting period we initiated experiments to assess the feasibility of assaying serum protein oxidation as a cumulative marker for the oxidation of macromolecules. Our rationale is based on the lack of information that exists about rates of formation and repair of oxidized adducts of DNA and lipid. Currently, it is difficult to estimate the timeframe that is represented in the measurement of these adducts, i.e. do they reflect acute exposure to various endogenous and exogenous factors or do they reflect cumulative exposures? We suspect that they may reflect steady sate conditions over relatively short time frames. What would be ideal is an index that reflects cumulative exposures. Some investigators have suggested that protein oxidation may be a accumulative index of oxidative status over time. As with DNA and lipids, many protein oxidation protects have been identified. The experiment reported analyzed levels of ortho-tyrosine and meta-tyrosine in serum protein using GC-MS. This work was done in collaboration

with Dr. Mark Duncan, Director of the University of Colorado mass spectrometry facility in the Department of Pharmacy. The following results were obtained.

Pearson correlation matrix

	OTYRPPM	MTYRPPM	LYOHDG	UREPGCT	URMDACT
OTYRPPM	1.000				
MTYRPPM	0.238	1.000			
LYOHDG	0.137	0.323	1.000		
UREPGCT	-0.028	0.620	0.862	1.000	
URMDACT	-0.541	0.418	0.599	0.738	1.000

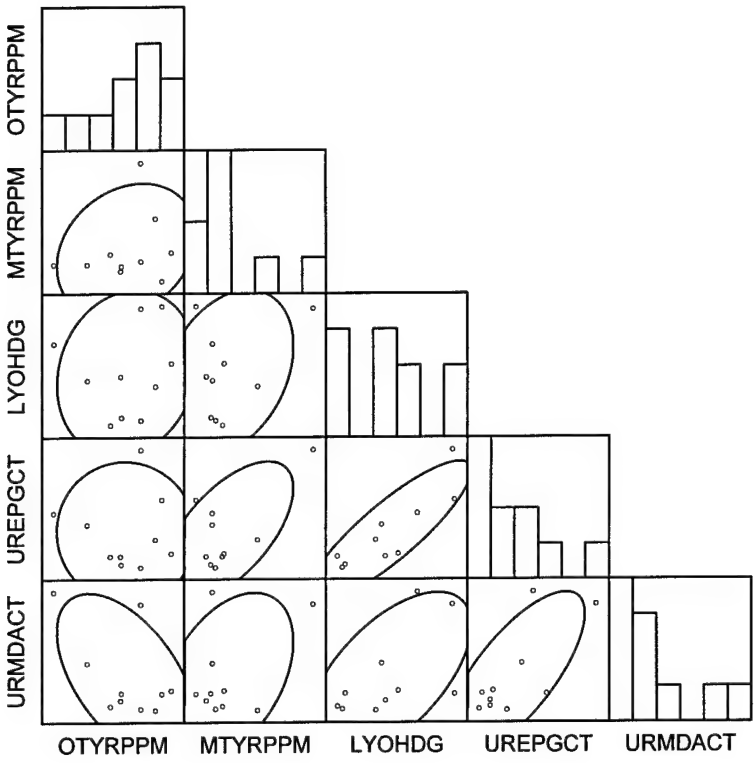
Abbreviations: Ortho-tyrosine, OTYRPPM; Meta-tyrosine, MTYRPPM; lymphocyte 8-hdroxydeoxyguanosine, LYOHDG; urinary 8-isoprostane F2-alpha, UREPGCT; urinary malondialdehyde, URMDACT.

Bartlett Chi-square statistic: 33.075 df=10 Prob= 0.000

Matrix of Bonferroni Probabilities

	OTYRPPM	MTYRPPM	LYOHDG	UREPGCT	URMDACT
OTYRPPM	0.000				
MTYRPPM	1.000	0.000			
LYOHDG	1.000	1.000	0.000		
UREPGCT	1.000	0.557	0.013	0.000	
URMDACT	1.000	1.000	0.674	0.149	0.000

Number of observations: 10



This study was intended as a feasibility study so the N was small. Therefore, most statistical tests have limited power. Nonetheless, positive associations were observed between both measures of DNA oxidation and serum protein oxidation. These results indicate the merit of additional experiments.

Key Research Accomplishments

- Obstacles to adherence to a recipe defined menu low or high in vegetable and fruit intake were identified. Strategies to overcome these barriers were developed, implemented, and tested. This has resulted in a new dietary intervention tool, i.e. a new cook book.
- The determination of 8-OHdG in urine by use of an ELISA assay was found to be unreliable. The use of a multi-column HPLC approach with electrochemical detection gave less variable results.
- The assessment of oxidative DNA damage by several modifications of the comet assay was found to provide information that was correlationally independent of that provided by the assessment of lymphocyte 8-OHdG or urinary 8-isoprostane F2-alpha.
- It is feasible to measure serum protein oxidation and the values obtained correlated positively with a measure of lymphocyte DNA oxidation.

Reportable Outcomes

- Cookbooks were developed and tested .
- Supporting intervention materials were developed and tested.
- An alternative method of analysis of a urinary product of DNA oxidation was identified.
- Assessment of serum protein oxidation was shown to be feasible.

Conclusions During this reporting period work was completed on the development of the intervention cookbook and on further refinement of assays for oxidative endpoints. Recruitment has been completed for the first intervention group with the experiment scheduled to begin in September 2000.

References

1. Park,E.M., Shigenaga,M.K., Degan,P., Korn,T.S., Kitzler,J.W., Wehr,C.M., Kolachana,P., and Ames,B.N. (1992) Assay of excised oxidative DNA lesions: isolation of 8-oxoguanine and its nucleoside derivatives from biological fluids with a monoclonal antibody column. *Proc.Natl.Acad.Sci U.S A.*, **89**, 3375-3379.

2. Halliwell,B. (1999) Establishing the significance and optimal intake of dietary antioxidants: the biomarker concept. *Nutr.Rev.*, **57**, 104-113.

3. Halliwell,B. (1998) Can oxidative DNA damage be used as a biomarker of cancer risk in humans? Problems, resolutions and preliminary results from nutritional supplementation studies. *Free Radic.Res.*, **29**, 469-486.

4. Inoue,M., Kamiya,H., Fujikawa,K., Ootsuyama,Y., Murata-Kamiya,N., Osaki,T., Yasumoto,K., and Kasai,H. (1998) Induction of chromosomal gene mutations in Escherichia coli by direct incorporation of oxidatively damaged nucleotides. New evaluation method for mutagenesis by damaged DNA precursors in vivo. *J.Biol.Chem.*, **273**, 11069-11074.

5. Suzuki,M., Matsui,K., Yamada,M., Kasai,H., Sofuni,T., and Nohmi,T. (1997) Construction of mutants of Salmonella typhimurium deficient in 8- hydroxyguanine DNA glycosylase and their sensitivities to oxidative mutagens and nitro compounds. *Mutat.Res.*, **393**, 233-246.

6. Demple,B. and Harrison,L. (1994) Repair of oxidative damage to DNA: enzymology and biology. *Annu.Rev.Biochem.*, **63:915-48**, 915-948.

7. Bessho,T., Tano,K., Kasai,H., Ohtsuka,E., and Nishimura,S. (1993) Evidence for two DNA repair enzymes for 8-hydroxyguanine (7,8-dihydro-8- oxoguanine) in human cells. *J.Biol.Chem.*, **268**, 19416-19421.

8. Reardon,J.T., Bessho,T., Kung,H.C., Bolton,P.H., and Sancar,A. (1997) In vitro repair of oxidative DNA damage by human nucleotide excision repair system: possible explanation for neurodegeneration in xeroderma pigmentosum patients. *Proc.Natl.Acad.Sci.U.S.A*, **94**, 9463-9468.

9. Klungland,A., Rosewell,I., Hollenbach,S., Larsen,E., Daly,G., Epe,B., Seeberg,E., Lindahl,T., and Barnes,D.E. (1999) Accumulation of premutagenic DNA lesions in mice defective in removal of oxidative base damage. *Proc.Natl.Acad.Sci.U.S.A*, **96**, 13300-13305.

10. Dianov,G., Bischoff,C., Piotrowski,J., and Bohr,V.A. (1998) Repair pathways for processing of 8-oxoguanine in DNA by mammalian cell extracts [In Process Citation]. *J.Biol.Chem.*, **273**, 33811-33816.

11. Le Page,F., Kwoh,E.E., Avrutskaya,A., Gentil,A., Leadon,S.A., Sarasin,A., and Cooper,P.K. (2000) Transcription-coupled repair of 8-oxoguanine: requirement for XPG, TFIIH, and CSB and implications for Cockayne syndrome. *Cell*, **101**, 159-171.

12. Prieme',H., Loft,S., Cutler,R.G., and Poulsen,H.E. (1996) Measurement of oxidative stress in humans: Evaluation of a commercially available ELISA assay. In Kumpulainin JT (ed.) *Natural antioxidants and food quality in atherosclerosis and cancer prevention.* Royal Society of Chemistry, Cambridge, UK, pp 78-82.

13. Haegele,A.D., Gillette,C., O'Neill,C., Wolfe,P., Heimendinger,J., Sedlacek,S., and Thompson,H.J. (2000) Plasma xanthophyll carotenoids correlate inversely with indices of oxidative DNA damage and lipid peroxidation. *Cancer Epidemiol.Biomarkers Prev.*, **9**, 421-425.

14. Bogdanov,M.B., Beal,M.F., McCabe,D.R., Griffin,R.M., and Matson,W.R. (1999) A carbon column-based liquid chromatography electrochemical approach to routine 8-hydroxy-2'-deoxyguanosine measurements in urine and other biologic matrices: a one-year evaluation of methods. *Free Radic.Biol.Med.*, **27**, 647-666.

Appendix A
Example of the format of version 1 of the intervention cook book

DAY 1

BREAKFAST

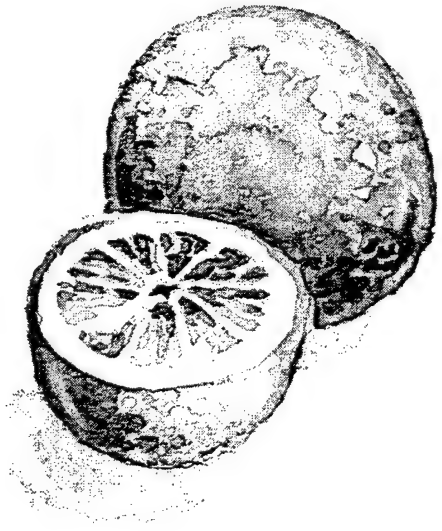
1/3 cup	100% orange juice
1-3/4 cup	Healthy Choice Toasted Brown Sugar Squares cereal
1 cup	fat-free milk

LUNCH

LYCOPENE SOUP
Prepared by Wild Oats Markets

Portion you must consume:
Lycopene Soup = 1 container
Bread = 2 slices
Butter = 2 tsp.

2 slices	100% whole wheat bread
2 tsp.	butter



DAY 1

BREAKFAST

ITEM	AMOUNT	% EATEN	COMMENTS
JUICE			
CEREAL			
MILK			

LUNCH

ITEM	MEASURE	% EATEN	COMMENTS
SOUP			
BREAD			
BUTTER			

DAY 1

DINNER

TERIYAKI CHICKEN

1 lb.	chicken breast, boneless, skinless, raw
	Teriyaki sauce (to cover)

Marinate chicken in the Teriyaki sauce for 2-4 hours in the refrigerator. Remove chicken from the marinade and place in a casserole dish; cover the casserole dish and bake at 350° F for 35-45 minutes, or until done. Uncover for the last 10 minutes to allow chicken to brown. Serves 3.

Portion you must consume:
Teriyaki Chicken = 1/3 of recipe
Baked Potato = entire recipe
milk = 1 cup
bread = 2 slices
butter = 2 tsp.

BAKED POTATO

3/4 medium	potato
2 tsp.	butter

Wash potato well. Puncture with a fork. Place on a microwave-safe dish. Microwave on high for 8-10 minutes or until cooked through. Mix with butter. Serves 1.

1 cup	fat-free milk
2 slices	100% whole wheat bread
2 tsp.	butter

DINNER

ITEM	AMOUNT	% EATEN	COMMENTS
CHICKEN			
POTATO			
BUTTER			
MILK			
BREAD			
BUTTER			

Appendix B
Dietary intervention evaluation instrument

Please feel free to make additional comments on any question.

1. How many servings of fruits and vegetables do you think a person should eat each day for good health?

0 ❑ None
1 ❑ 1
2 θ 2
3 θ 3
4 ❑ 4
5 ❑ 5
6 ❑ 6
7 ❑ 7
8 ❑ 8
9 ❑ 9
10 ❑ 10
11 ❑ 11 or more

For questions 2 and 3, a serving is defined as:

- a medium piece of fruit
- ¼ cup of dried fruit or vegetable
- ½ cup cooked or raw fruit or vegetable
- ½ cup dried peas or beans
- 1 cup leafy greens

2. How confident are you that you can include 2 servings of fruit at breakfast every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

3. How confident are you that you can include 5 – 9 servings of fruits and vegetables in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

4. How confident are you that you could eat more fruits and vegetables every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident
6 θ I already eat enough fruits and vegetables

5. How confident are you that you can include whole grains (brown rice, 100% whole-wheat toast) in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

6. How confident are you that you can include leafy greens in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

7. Within your daily routine, how convenient was it to go to Wild Oats to pick up your entrees?

1 ❑ Very inconvenient
2 ❑ Inconvenient
3θ Neither
4 ❑ Convenient
5 ❑ Very convenient

8. Once you were at the Wild Oats deli, how easy was the pick-up procedure?

1 ❑ Very difficult
2 ❑ Difficult
3θ Neither
4 ❑ Easy
5 ❑ Very easy

9. How well did the Wild Oats entrees reheat?

1 ❑ Very poorly
2 ❑ Poorly
3θ Neither
4 ❑ Well
5 ❑ Very well

10.How well did the Wild Oats packaging (containers, lids) remain intact?

1 ❑ Very poorly
2 ❑ Poorly
3θ Neither
4 ❑ Well
5 ❑ Very well

11. How easy was it for you to store the food from Wild Oats in your freezer/refrigerator?

1 ❑ Very difficult
2 ❑ Difficult
3θ Neither
4 ❑ Easy
5 ❑ Very easy

12. How easy was it for you to store the food purchased at the grocery store?

1 ❑ Very difficult
2 ❑ Difficult
3θ Neither
4 ❑ Easy
5 ❑ Very easy

13. Did you go out of town during the study?

1 ❑ Yes
2 ❑ No (go to question 14)

13a. Did you follow the diet or substitute foods while you were out of town?

1 ❑ Followed the diet (answer question 13b, then skip to question 14)
2 ❑ Substituted foods (skip to question 13c)
3θ Both

13b. How easy was it to follow the diet while you were out of town?

1 ❑ Very difficult
2 ❑ Difficult
3θ Neither
4 ❑ Easy
5 ❑ Very easy

13c. Did you receive assistance in making substitutions to your diet from the study staff while you were out of town?

1 ❑ Yes
2 ❑ No

13d. How easy was it to make substitutions to your diet while you were out of town?

1 ❑ Very difficult
2 ❑ Difficult
3θ Neither
4 ❑ Easy
5 ❑ Very easy

14. Did you attend a special event (e.g. wedding, play, banquet) during the study?

1 ❑ Yes
2 ❑ No (go to question 15)

14a. Did you follow the diet or substitute foods when you attended the special event?

1 ❑ Followed the diet (answer question 14b, then skip to question 15)
2 ❑ Substituted foods (skip to question 14c)
3θ Both

14b. How easy was it to follow the diet when you attended the special event?

1 ❑ Very difficult
2 ❑ Difficult
3θ Neither
4 ❑ Easy
5 ❑ Very easy

14c. Did you receive assistance in making substitutions to your diet from the study staff when you attended the special event?

1 ❑ Yes
2 ❑ No

14d. How easy was it to make substitutions to your diet when you attended the special event?

1 ❑ Very difficult
2 ❑ Difficult
3θ Neither
4 ❑ Easy
5 ❑ Very easy

15. Could you stay on the intervention diet for 8 weeks?

1 ❑ No
2 ❑ Yes

If you answered no to question 15, what changes could we make that would help you stay on the intervention diet for 8 weeks?

16. If you were allowed 2 non-study meals each week and allowed 1 alcoholic beverage per day could you stay on the intervention diet for 8 weeks?

1 ❑ No
2 ❑ Yes

17. Would holidays falling during the 8 week study prevent you from wanting to participate?

1 ❑ No
2 ❑ Yes

If you answered yes to question 17, which holidays in particular would prevent you from participating?

18. On a scale of 1 to 5, with 1 being "very unlikely" and 5 being "very likely", please indicate how likely you would be to follow the diet for 8 weeks if it included the following foods:

Eggs:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Waffles:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Pancakes:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Beef:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Luncheon Meats:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Desserts:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Chocolate/Candy:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Alcohol:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑

Please comment on any additional foods that would help you to follow the diet for 8 weeks:

19. Please tell us things you liked and disliked about the first meeting on March 18.

20. Please tell us what would make the intervention easier for you to follow.

21. Overall, please tell us what you liked about this study.

22. Overall, please tell us what you disliked about this study.

23. Do you plan to make any changes in your dietary habits as a result of participating in this study?

1 ❑ No
2 ❑ Yes

If you answered yes to question 23, what do you plan to do?

If you were in Group A, answer questions 24 and 25. If you were in Group B, answer questions 26 and 27.

24. On a scale of 1 to 5, with 1 being "disliked very much" and 5 being "liked very much", please indicate how you liked the following foods prepared by Wild Oats:

Lycopene Soup:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Broccoli-Mushroom Pasta:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Tofu-Parsley Dressing:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Springtime Vegetable Stew:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Orange-Glazed Sweet Potatoes:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Cauliflower Linguine:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Orange-Poppy Seed Dressing:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Lentil Stew:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Garlic Vegetable Stir Fry:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Vegetarian Chili:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Pasta and Beans with Basil:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Mexican Stew:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Steamed Carrot, Cauliflower and Apricots:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑

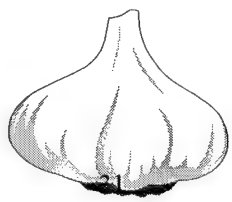
Comments: _____

25.On a scale of 1 to 5, with 1 being "disliked very much" and 5 being "liked very much", please indicate how you liked the following recipes from the cookbook:

Wilted Spinach Pasta Peanut Salad:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Banana-Orange Smoothie:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Teriyaki Chicken:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Pasta and Broccoli Salad:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Banana-Raspberry Smoothie:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Pepper Crusted Tuna:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Blueberry-Strawberry Smoothie:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Spinach Salad with Mandarin Orange:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Cantaloupe-Banana Smoothie:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Shrimp-Avocado Salad:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Broccoli and Cauliflower Baked Potato:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Cottage Cheese Salad:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Cucumber-Green Pepper Salad:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Tuna Salad Sandwich:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑

Comments: _____

Thank you!



26. On a scale of 1 to 5, with 1 being "disliked very much" and 5 being "liked very much", please indicate how you liked the following foods prepared by Wild Oats:

Lycopene Soup:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Tofu-Broccoli Stir Fry:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Pasta with Herb-Cheese Sauce:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Carrots with Cauliflower:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Pasta with White Bean Sauce:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Risotto de Napoli:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Macaroni and Cheese a la Moutarde:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Cajun Rice and Red Beans:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑

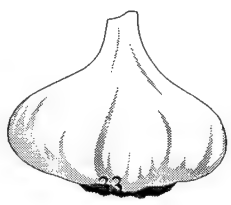
Comments: _____

27.On a scale of 1 to 5, with 1 being "disliked very much" and 5 being "liked very much", please indicate how you liked the following recipes from the cookbook:

Teriyaki Chicken:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Avocado-Pasta Shrimp Salad:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Broiled Salmon:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Roasted Quesadilla Ole:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Pepper Crusted Tuna:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Spaghetti:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑
Tuna Salad Sandwich:	1 ❑	2 ❑	3 θ	4 ❑	5 ❑

Comments: _____

Thank you!



For participants who have participated in a previous Cuisine for Cancer Prevention study:

1. In the current study you were provided the Informed Consent document ahead of time and you were given an opportunity to ask questions about the study individually. In the previous study we discussed the Informed Consent document in a group and then you were given a chance to ask individual questions. Which approach did you prefer?

   1 ❑ Preferred individual discussion only
   2 ❑ Preferred group discussion followed by individual discussion

2. The current study required 2 meetings, while the previous studies required 3 meetings. Did you prefer having 2 meetings or 3 meetings?

   1 ❑ Preferred 2 meetings
   2 ❑ Preferred 3 meetings

3. In the current study we did meet on the Saturday that falls in the middle of the 2 week diet. In previous studies, we met on that Saturday to answer questions about the diet and/or share why you chose to participate in the study. Please tell us how you felt about not having the 2nd meeting during this intervention.

4. The current study required you to measure your food in cups, while the previous study required you to weigh your food. Did you prefer measuring your food or weighing your food?

   1 ❑ Preferred measuring the food
   2 ❑ Preferred weighing the food

5. Please tell us how you feel about measuring food vs. weighing food.

6. How convenient do you feel this study was compared to the previous cuisine studies that you have participated in?

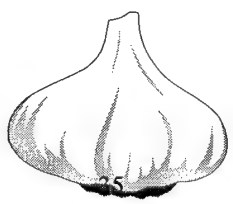
1 ❑ A lot more inconvenient
2 ❑ Somewhat more inconvenient
3θ The same as previous study(ies)
4 ❑ Somewhat more convenient
5 ❑ A lot more convenient

7. Overall, did you prefer this study with food prepared by Wild Oats or previous studies with foods prepared at home?

1 ❑ Preferred this study
2 ❑ Preferred previous study(ies)

Please tell us the reasons for your preference above.

Thank you!



Appendix C
Example of final cookbook format

CUISINE for Cancer PREVENTION

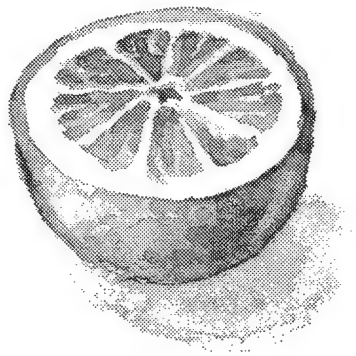
AMC
Cancer Research Center

A national not-for-profit research institute dedicated to
the prevention of cancer and other chronic diseases.

1600 Pierce Street • Denver, CO • 80214 • www.amc.org
1-800-525-3777 Cancer Information and Counseling Line
1-800-321-1557 Research and Foundation Information

DAY 1

BREAKFAST



You must eat:....1 juice exchange, **AND**
1 dairy/soy exchange, **AND**
1 bread/cereal exchange

Juice:	1/3 cup	orange juice, **OR**
	1/3 cup	grapefruit juice, **OR**
	1/3 cup	tangerine juice
Dairy/Soy: ..	1 cup	fat-free milk, **OR**
	1 cup	fat-free fortified soy milk, **OR**
	1 cup	nonfat yogurt (any flavor)
Bread/Cereal: ..	1 medium	frozen waffle (4" diameter) +2 tsp. butter +1 Tbsp. syrup, **OR**
	1/2 large	plain bagel (4" diameter) +2 Tbsp. cream cheese, **OR**
	1 medium	English muffin (3-1/2" diameter) +1 Tbsp. peanut butter, **OR**
	2 medium	slices white bread +2 tsp. butter +2 tsp. jelly (any flavor), **OR**
	2/3 cup	Post Morning Traditions Great Grains Crunchy Pecan cereal, **OR**
	1/2 cup	Quaker Sun Country Granola with Almonds cereal, **OR**
	1/2 cup	Kellogg's Cracklin' Oat Bran cereal

BREAKFAST

EXCHANGE	ITEM	AMOUNT	% EATEN	COMMENTS
JUICE EXCHANGE	Orange ❑ Grapefruit ❑ Tangerine ❑			
DAIRY/SOY EXCHANGE	Milk ❑ Soy Milk ❑ Yogurt ❑			
BREAD/CEREAL EXCHANGE	Waffle ❑ Butter ❑ Syrup ❑			
	Bagel ❑ Cream Cheese ❑			
	English Muffin ❑ Butter ❑			
	Bread ❑ Butter ❑			
	Great Grains Cereal ❑			
	Granola Cereal ❑			
	Oat Bran Cereal ❑			

DAY 1

LUNCH

You must eat:....Lycopene Soup, **AND**
1 bread exchange

LYCOPENE SOUP Prepared by Wild Oats Markets

Bread: .....2 medium slices white bread + 2 tsp. butter, **OR**
12 Keebler Club crackers, **OR**
1/2 large plain bagel (4" diameter) +2 tsp. butter

DINNER

You must eat:....1 meat exchange, **AND**
1 vegetable exchange, **AND**
1 dairy exchange, **AND**
1 bread/pasta/rice exchange

Meat: .....1 lb. chicken breast, boneless, skinless, raw, **OR**
1 lb. salmon fillet, raw, **OR**
1 lb. sirloin steak, raw

Teriyaki sauce (optional)

If desired, marinate the chicken, salmon, or sirloin steak in teriyaki sauce for 2-4 hours in the refrigerator. Remove from the marinade and cook as follows:

Note: This entrée is served again for dinner on Day 4 and Day 11. You may wish to freeze these portions or you can make the recipe again for those days.

DAY 1

LUNCH

EXCHANGE	ITEM	AMOUNT	% EATEN	COMMENTS
SOUP	Soup ❑			
BREAD EXCHANGE	Bread ❑ Butter ❑			
	Crackers ❑			
	Bagel ❑ Butter ❑			

DINNER

EXCHANGE	ITEM	AMOUNT	% EATEN	COMMENTS
MEAT EXCHANGE	Chicken ❑ Salmon ❑ Steak ❑			
MARINADE (OPTIONAL)	Teriyaki ❑			

DINNER (cont.)

Chicken: bake in covered casserole dish at 350°F for 35-45 minutes, or until done. Uncover for the last 10 minutes to allow chicken to brown. Serves 3.

Salmon: broil for 5-8 minutes, or until done. Serves 3.

Sirloin Steak: broil for 5-8 minutes per side, or until done. Serves 3.

Vegetable:	1/2	medium potato, **OR**
	1/2	medium eggplant
	2 tsp.	butter

Cook as follows:

Potato: Wash well; puncture with a fork. Microwave on high for 8-10 minutes or until cooked through. Mix with butter. Serves 1.

Eggplant: Slice eggplant into 1/2" strips. Grill or broil until tender. Toss with butter. Serves 1.

Dairy/Soy:	1 cup	fat-free fortified soy milk, **OR**
	1 cup	fat-free milk, **OR**
	1 cup	nonfat yogurt (any flavor)

Bread:	2 medium	slices white bread, **OR**
	1 cup	pasta (any type), **OR**
	1 cup	white rice, **OR**
	2 tsp.	butter

DINNER (cont.)

EXCHANGE	ITEM	AMOUNT	% EATEN	COMMENTS
POTATO/ EGGPLANT EXCHANGE	Potato ❑ Eggplant ❑			
BUTTER	Butter ❑			
MILK/SOY EXCHANGE	Soy Milk ❑ Milk ❑ Yogurt ❑			
BREAD/PASTA/ RICE EXCHANGE	Bread ❑ Pasta ❑ Rice ❑			
BUTTER	Butter ❑			

Appendix D
Other intervention materials

1. What is the highest level of education you have completed?

1 ❑ Grade school
2 ❑ Some high school
3 θ High school graduate
4 θ Some college
5 θ College graduate (4-year degree)
6 θ Post-graduate

2. Which of these categories best describes you?

10 ❑ Asian or Pacific Islander
20 ❑ Black or African American
30 ❑ Hispanic
40 θ American Indian or Alaskan Native
50 θ White, Non-Hispanic
60 ❑ Other

3. Do you live with a spouse or significant other? 1 ❑ yes 2 ❑ no

4. Do you live with adults other than a spouse or significant other, not including adult children? 1 ❑ yes 2 ❑ no

5. Do you live with children under 18 years of age? 1 ❑ yes 2 ❑ no

6. Do you live with children 18 years of age or older? 1 ❑ yes 2 ❑ no

7. How much responsibility do you have for preparing meals?

0 ❑ None
1 ❑ Hardly any
2 θ Some
3 θ Most
4 θ All

8. How many servings of fruits and vegetables do you think a person should eat each day for good health?

0	❑	None	6	❑	6
1	❑	1	7	❑	7
2	θ	2	8	❑	8
3	θ	3	9	❑	9
4	❑	4	10	❑	10
5	❑	5	11	❑	11 or more

9. It is difficult to get fruits and vegetables when I eat out in restaurants.

1	❑	Strongly Disagree
2	❑	Disagree
3	❑	Neither agree nor disagree
4	❑	Agree
5	❑	Strongly Agree

10. I don't know how to prepare fruits and vegetables.

1	❑	Strongly Disagree
2	❑	Disagree
3	❑	Neither agree nor disagree
4	❑	Agree
5	❑	Strongly Agree

11. Over the past month, about how often did you drink 100% orange juice or grapefruit juice?

1	❑	Never (less than once a month)	6	❑	1 time per day
2	❑	1-3 times per month	7	❑	2 times per day
3	θ	1-2 times per week	8	❑	3 times per day
4	❑	3-4 times per week	9	❑	4 times per day
5	θ	5-6 times per week	10	θ	5 or more times per day

12. Over the past month, about how often did you drink 100% juice other than orange or grapefruit juices? Do not count fruit juices like Hi-C or Hawaiian Punch that are only part juice.

1	❑	Never (less than once a month)	6	❑	1 time per day
2	❑	1-3 times per month	7	❑	2 times per day
3	θ	1-2 times per week	8	❑	3 times per day
4	❑	3-4 times per week	9	❑	4 times per day
5	θ	5-6 times per week	10	θ	5 or more times per day

13. Over the past month, about how often did you eat green salad with or without other vegetables?

1 ❑ Never (less than once a month)
2 ❑ 1-3 times per month
3θ 1-2 times per week
4 ❑ 3-4 times per week
5 θ 5-6 times per week
6 ❑ 1 time per day
7 ❑ 2 times per day
8 ❑ 3 times per day
9 ❑ 4 times per day
10 θ 5 or more times per day

14. Over the past month, about how often did you eat french fries or fried potatoes?

1 ❑ Never (less than once a month)
2 ❑ 1-3 times per month
3θ 1-2 times per week
4 ❑ 3-4 times per week
5 θ 5-6 times per week
6 ❑ 1 time per day
7 ❑ 2 times per day
8 ❑ 3 times per day
9 ❑ 4 times per day
10 θ 5 or more times per day

15. Over the past month, about how often did you eat baked, boiled, or mashed potatoes?

1 ❑ Never (less than once a month)
2 ❑ 1-3 times per month
3θ 1-2 times per week
4 ❑ 3-4 times per week
5 θ 5-6 times per week
6 ❑ 1 time per day
7 ❑ 2 times per day
8 ❑ 3 times per day
9 ❑ 4 times per day
10 θ 5 or more times per day

For the following questions, a serving is defined as:

- a medium piece of fruit
- ¼ cup of dried fruit or vegetable
- ½ cup cooked or raw fruit or vegetable
- ½ cup dried peas or beans

16. Over the past month, about how many servings of vegetables did you eat not counting salad or potatoes?

1 ❑ None (less than one per month)
2 ❑ 1-3 per month
3θ 1-2 per week
4 ❑ 3-4 per week
5 θ 5-6 per week
6 ❑ 1 per day
7 ❑ 2 per day
8 ❑ 3 per day
9 ❑ 4 per day
10 θ 5 or more per day

17. Over the past month, about how many servings of fruit did you eat, not counting juices?

1 ❑ None (less than one per month)
2 ❑ 1-3 per month
3θ 1-2 per week
4 ❑ 3-4 per week
5 θ 5-6 per week
6 ❑ 1 per day
7 ❑ 2 per day
8 ❑ 3 per day
9 ❑ 4 per day
10 θ 5 or more per day

18. Over the past month, about how many servings of meat, poultry, fish, dry beans, eggs and nuts did you eat? Examples of a serving include 3 ounces of cooked meat, poultry or fish (3 ounces is about the size and thickness of a deck of cards); 1½ cup cooked dried beans; 6 tablespoons peanut butter; 3 eggs; and 1 cup nuts.

1 ❑ None (less than one per month)
2 ❑ 1-3 per month
3θ 1-2 per week
4 ❑ 3-4 per week
5 θ 5-6 per week
6 ❑ 1 per day
7 ❑ 2 per day
8 ❑ 3 per day
9 ❑ 4 per day
10 θ 5 or more per day

19. Over the past month, about how many servings of milk, yogurt and cheese did you eat? Examples of a serving include 1 cup of milk, 1 cup of yogurt, and 1½ ounces of cheese.

1 ❑ None (less than one per month)
2 ❑ 1-3 per month
3θ 1-2 per week
4 ❑ 3-4 per week
5 θ 5-6 per week
6 ❑ 1 per day
7 ❑ 2 per day
8 ❑ 3 per day
9 ❑ 4 per day
10 θ 5 or more per day

20. Over the past month, about how many servings of bread, cereal, rice and pasta did you eat? Examples of a serving include 1 slice of bread; ½ cup cooked cereal, rice, or pasta; 1 cup ready-to-eat cereal; and 1 small roll, biscuit or muffin.

1 ❑ None (less than one per month)
2 ❑ 1-3 per month
3θ 1-2 per week
4 ❑ 3-4 per week
5 θ 5-6 per week
6 ❑ 1-3 per day
7 ❑ 4-6 per day
8 ❑ 7-9 per day
9 ❑ 9-11 per day
10 θ 12 or more per day

21. How confident are you that you can include 2 servings of fruit at breakfast every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

22. How confident are you that you can include 5 – 9 servings of fruits and vegetables in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

23. How confident are you that you could eat more fruits and vegetables every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident
6 θ I already eat enough fruits and vegetables

24. How confident are you that you can include whole grains (brown rice, 100% whole-wheat toast) in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

25. How confident are you that you can include leafy greens in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

26. How much does your family encourage you to eat fruits and vegetables?

1 ❑ A great deal
2 ❑ Somewhat
3 ❑ Not much
4 ❑ Not at all
997 ❑ Not applicable

27. How much do your friends encourage you to eat fruits and vegetables?

1 ❑ A great deal
2 ❑ Somewhat
3 ❑ Not much
4 ❑ Not at all
997 ❑ Not applicable

28. Does your family plan to follow the study diet while you are participating in the study?

1 ❑ Yes 2 ❑ No 997 ❑ Not applicable

1. How many servings of fruits and vegetables do you think a person should eat each day for good health?

0	❑	None	6	❑	6
1	❑	1	7	❑	7
2	θ	2	8	❑	8
3	θ	3	9	❑	9
4	❑	4	10	❑	10
5	❑	5	11	❑	11 or more

2. Over the past month, about how often did you drink 100% orange juice or grapefruit juice?

1	❑	Never (less than once a month)	6	❑	1 time per day
2	❑	1-3 times per month	7	❑	2 times per day
3	θ	1-2 times per week	8	❑	3 times per day
4	❑	3-4 times per week	9	❑	4 times per day
5	θ	5-6 times per week	10	θ	5 or more times per day

3. Over the past month, about how often did you drink 100% juice other than orange or grapefruit juices? Do not count fruit juices like Hi-C or Hawaiian Punch that are only part juice.

1	❑	Never (less than once a month)	6	❑	1 time per day
2	❑	1-3 times per month	7	❑	2 times per day
3	θ	1-2 times per week	8	❑	3 times per day
4	❑	3-4 times per week	9	❑	4 times per day
5	θ	5-6 times per week	10	θ	5 or more times per day

4. Over the past month, about how often did you eat green salad with or without other vegetables?

1	❑	Never (less than once a month)	6	❑	1 time per day
2	❑	1-3 times per month	7	❑	2 times per day
3	θ	1-2 times per week	8	❑	3 times per day
4	❑	3-4 times per week	9	❑	4 times per day
5	θ	5-6 times per week	10	θ	5 or more times per day

5. Over the past month, about how often did you eat french fries or fried potatoes?

1	❑	Never (less than once a month)	6	❑	1 time per day
2	❑	1-3 times per month	7	❑	2 times per day
3	θ	1-2 times per week	8	❑	3 times per day
4	❑	3-4 times per week	9	❑	4 times per day
5	θ	5-6 times per week	10	θ	5 or more times per day

6. Over the past month, about how often did you eat baked, boiled, or mashed potatoes?

1	❑	Never (less than once a month)	6	❑	1 time per day
2	❑	1-3 times per month	7	❑	2 times per day
3	θ	1-2 times per week	8	❑	3 times per day
4	❑	3-4 times per week	9	❑	4 times per day
5	θ	5-6 times per week	10	θ	5 or more times per day

For the following questions, a serving is defined as:

- a medium piece of fruit
- ¼ cup of dried fruit or vegetable
- ½ cup cooked or raw fruit or vegetable
- ½ cup dried peas or beans

7. Over the past month, about how many servings of vegetables did you eat not counting salad or potatoes?

1	❑	None (less than one per month)	6	❑	1 per day
2	❑	1-3 per month	7	❑	2 per day
3	θ	1-2 per week	8	❑	3 per day
4	❑	3-4 per week	9	❑	4 per day
5	θ	5-6 per week	10	θ	5 or more per day

8. Over the past month, about how many servings of fruit did you eat, not counting juices?

1	❑	None (less than one per month)	6	❑	1 per day
2	❑	1-3 per month	7	❑	2 per day
3	θ	1-2 per week	8	❑	3 per day
4	❑	3-4 per week	9	❑	4 per day
5	θ	5-6 per week	10	θ	5 or more per day

9. Over the past month, about how many servings of meat, poultry, fish, dry beans, eggs and nuts did you eat? Examples of a serving include 3 ounces of cooked meat, poultry or fish (3 ounces is about the size and thickness of a deck of cards); 1½ cup cooked dried beans; 6 tablespoons peanut butter; 3 eggs; and 1 cup nuts.

1	❑	None (less than one per month)	6	❑	1 per day
2	❑	1-3 per month	7	❑	2 per day
3	θ	1-2 per week	8	❑	3 per day
4	❑	3-4 per week	9	❑	4 per day
5	θ	5-6 per week	10	θ	5 or more per day

10. Over the past month, about how many servings of milk, yogurt and cheese did you eat? Examples of a serving include 1 cup of milk, 1 cup of yogurt, and 1½ ounces of cheese.

1 ❑ None (less than one per month)
2 ❑ 1-3 per month
3θ 1-2 per week
4 ❑ 3-4 per week
5 θ 5-6 per week
6 ❑ 1 per day
7 ❑ 2 per day
8 ❑ 3 per day
9 ❑ 4 per day
10 θ 5 or more per day

11. Over the past month, about how many servings of bread, cereal, rice and pasta did you eat? Examples of a serving include 1 slice of bread; ½ cup cooked cereal, rice, or pasta; 1 cup ready-to-eat cereal; and 1 small roll, biscuit or muffin.

1 ❑ None (less than one per month)
2 ❑ 1-3 per month
3θ 1-2 per week
4 ❑ 3-4 per week
5 θ 5-6 per week
6 ❑ 1-3 per day
7 ❑ 4-6 per day
8 ❑ 7-9 per day
9 ❑ 9-11 per day
10 θ 12 or more per day

12. How confident are you that you can include 2 servings of fruit at breakfast every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

13. How confident are you that you can include 5 – 9 servings of fruits and vegetables in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

14. How confident are you that you could eat more fruits and vegetables every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident
6 θ I already eat enough fruits and vegetables

15. How confident are you that you can include whole grains (brown rice, 100% whole-wheat toast) in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

16. How confident are you that you can include leafy greens in your diet every day?

1 ❑ Not at all confident
2 ❑ Not confident
3θ Neither
4 ❑ Confident
5 ❑ Completely confident

17. How much does your family encourage you to eat fruits and vegetables?

1 ❑ A great deal
2 ❑ Somewhat
3 ❑ Not much
4 ❑ Not at all
997 ❑ Not applicable

18. How much do your friends encourage you to eat fruits and vegetables?

1 ❑ A great deal
2 ❑ Somewhat
3 ❑ Not much
4 ❑ Not at all
997 ❑ Not applicable

19. Have you made any changes in your diet since you completed the study diet?

1 ❑ Yes
2 ❑ No

If you answered yes to question 19, what types of changes have you made?